Development of an Oligonucleotide Array for Direct Detection of Fungi in Sputum Samples from Patients with Cystic Fibrosis

Titre	Development of an Oligonucleotide Array for Direct Detection of Fungi in Sputum Samples from Patients with Cystic Fibrosis
Type de publication	Article de revue
Auteur	Bouchara, Jean-Philippe [1], Hsieh, Hsin Yi [2], Croquefer, Sabine [3], Barton, Richard [4], Apaire-Marchais, Véronique [5], Pihet, Marc [6], Chang, Tsung Chain [7]
Editeur	American Society for Microbiology
Type	Article scientifique dans une revue à comité de lecture
Année	2009
Langue	Anglais
Date	2009/01/01
Numéro	1
Pagination	142 - 152
Volume	47
Titre de la revue	Journal of Clinical Microbiology
ISSN	1098-660X
Résumé en anglais	Cystic fibrosis (CF) is the most common inherited genetic disease in Caucasian populations. Besides bacteria, many species of fungi may colonize the respiratory tract of these patients, sometimes leading to true respiratory infections. In this study, an oligonucleotide array capable of identifying 20 fungal species was developed to directly detect fungi in the sputum samples of CF patients. Species-specific oligonucleotide probes were designed from the internal transcribed spacer (ITS) regions of the rRNA operon and immobilized on a nylon membrane. The fungal ITS regions were amplified by PCR and hybridized to the array for species identification. The array was validated by testing 182 target strains (strains which we aimed to identify) and 141 nontarget strains (135 species), and a sensitivity of 100% and a specificity of 99.2% were obtained. The validated array was then used for direct detection of fungi in 57 sputum samples from 39 CF patients, and the results were compared to those obtained by culture. For 16 sputum samples, the results obtained by the array corresponded with those obtained by culture. For 33 samples, the array detected more fungal species than culture did, while the reverse was found for eight samples. The accuracy of the array for fungal detection in sputum samples was confirmed (or partially confirmed) in some samples by cloning and resequencing the amplified ITS fragments. The present array is a useful tool for both the simultaneous detection of multiple fungal species present in the sputa of CF patients and the identification of fungi isolated from these patients.
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DOI	10.1128/JCM.01668-08 [9]
Lien vers le document	http://dx.doi.org/10.1128/JCM.01668-08 [9]

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